# Effects of selected insecticides on *Cotesia plutellae*, endoparasitoid of *Plutella xylostella*

M. HASEEB<sup>1,3</sup>, T.-X. LIU<sup>1,\*</sup> and W.A. JONES<sup>1,2</sup>

<sup>1</sup>Vegetable IPM Laboratory, Texas Agricultural Experiment Station, Texas A&M University, 2415 East Highway 83, Weslaco, TX 78596-8399, USA; <sup>2</sup>USDA, ARS, Kika de la Garza Subtropical Agricultural Research Center, Beneficial Insect Research Unit, 2413 E. Highway 83, Weslaco, TX 78596, USA; <sup>3</sup>Current address: Center for Biological Control, Florida A&M University, Tallahassee, FL 32307-4100, USA \*Author for correspondence; e-mail: tx-liu@tamu.edu

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Abstract. Effects of field dosages of selected insecticides to Cotesia plutellae (Kurdjumov) (Hymenoptera: Braconidae), a larval endoparasitoid of Plutella xylostella L. (Lepidoptera: Plutellidae), were investigated under laboratory conditions. Emergence of adult C. plutellae from insecticide-treated pupae was not significantly different from the control treatment. Contact toxicity to C. plutellae adults varied greatly among the insecticides in a paper residue contact bioassay. Three azadirachtin-based insecticides, Agroneem (4.8 mg a.i.liter<sup>-1</sup>), Neemix (20 mg a.i.liter<sup>-1</sup>) and Ecozin (20 mg ai.liter<sup>-1</sup>) caused 11.1, 16.7 and 5.6% adult mortality, respectively. Of four commercial Bacillus thuringiensis (Bt) insecticides examined (all at 1.2 mg a.i.liter<sup>-1</sup>), Crymax and Xentari had no effect on adult parasitoids, whereas Mattch caused 5.6% mortality, and Dipel caused 11.1% mortality. Indoxacarb (53 mg a.i.liter<sup>-1</sup>), λcyhalothrin (28 mg a.i.liter<sup>-1</sup>) and spinosad (53 mg a.i.liter<sup>-1</sup>) caused 100, 88.5 and 50% adult mortalities, respectively. Low adult mortality (0-5.6%) was recorded from ingestion of azadirachtin-based, Bt insecticides and indoxacarb, compared with 100% adult mortality in treatments of spinosad or λ-cyhalothrin. Compared with the water control, ingestion of azadirachtin-based insecticides significantly reduced parasitism by 50-57%, and Bt insecticides by 8-25%. However, ingestion of these insecticides did not affect longevity of male and female parasitoid adults with one exception; female longevity was significantly reduced in the indoxacarb treatment. Insecticide residues caused considerable mortality of C. plutellae adults, 39 and 44% mortality caused by 10 d old indoxacarb and λ-cyhalothrin, respectively, and 24 and 0% mortality caused by 7 and 10 d old residues of spinosad, respectively.

**Key words:** Cotesia plutellae, diamondback moth, insecticides, IPM, non-target effects, parasitoid, Plutella xylostella

#### Introduction

The diamondback moth, *Plutella xylostella* L. (Lepidoptera: Plutellidae) is one of the most destructive pests of cruciferous crops throughout the world.

In Southeast Asia, major outbreaks of *P. xylostella* can cause >90% crop losses (Verkerk and Wright, 1996). Historically, this pest typically remained below economic threshold levels in the United States and Europe by various means (Shelton et al., 1993). However, current trends are for increased use of insecticides (Shelton et al., 1993; Liu and Sparks, 1999). The main drawbacks of chemical control of *P. xylostella* include development of insecticide resistance, resurgence of the pest after insecticide application, and nonselective killing of natural enemies (Nemoto, 1986).

Cotesia plutellae (Kurdjumov) is a solitary larval endoparasitoid of *P. xylostella*. It is reported to be host-specific to *P. xylostella* (Verkerk and Wright, 1996), although there are a few records of it attacking other lepidopterous hosts, such as *Ocnogyma baetica* (Rambur) in Spain (Lipa et al., 1993), and *Autographa gamma* (L.) in Japan (Kaneko, 1993). In Japan, field parasitism of *P. xylostella* ranges from 18 to 83% (Haseeb et al., 2001). In South Africa, Waladde et al. (2001) found that *C. plutellae* was active year round, accounting for 30–50% parasitism. Field experiments using three cabbage cultivars, grown under pesticide-free conditions, demonstrated that *C. plutellae* achieved rates of parasitism as high as 90–95% for at least eight consecutive months. Johnson et al. (1988) reported that *C. plutellae* was the dominant parasitoid species on cabbage and other crucifer vegetables in Hawaii.

There is a recognized need for implementation of more sustainable integrated pest management (IPM) strategies with a greater emphasis on biologically-based tactics of crop protection and reduced reliance on broad spectrum insecticides. However, chemical insecticides usually play a major role in management of P. xylostella. Currently growers are facing serious threats from this pest, particularly due to insecticide resistance and ineffective biological control. Insecticides exert a wide variety of direct and indirect effects on natural enemies. Lethal effects are often expressed as acute or chronic mortality resulting from contact with or ingestion of insecticides. However, sublethal effects on natural enemies are typically chronic, and not obvious. These may be expressed as changes in life-history traits of insects, such as parasitism rate, longevity, egg viability, consumption rate, or behavior (Ruberson et al., 1998). Thus, chemical insecticides need to be correctly and selectively used to ensure sustainable crop protection and environmental stability (Jepson, 1989; Greathead, 1995; Haseeb et al., 2000; Haseeb, 2001). Broad-spectrum insecticides can often cause lethal or sublethal effects on beneficial species that may not only upset the optimal population density in the field but may also impose certain negative impacts on the species itself (Amano and Haseeb, 2001; Haseeb and Amano, 2002). The current study was designed to investigate certain lethal effects of currently-used insecticides on *C. plutellae* and their possible implications for the management of *P. xylostella* on cabbage.

#### Materials and methods

## Pest culture and parasitoid source

Plutella xylostella larvae were collected from an insecticide-free cabbage field at the Texas A&M University Agricultural Research and Extension Center, Weslaco, USA. P. xylostella larvae were reared on radish sprouts in plastic boxes (24 cm  $\times$  15 cm  $\times$  12 cm) under laboratory conditions at 25  $\pm$  2 °C, 60  $\pm$  10% RH and a photoperiod of 16L:8D (Haseeb et al., 2000). After cleaning and softening in warm water and being sterilized with 1% benomyl (Benlate, 50% DF, Du Pont, Wilmington, DE) to prevent growth of fungal mold, about 40 g of radish sprouts were placed in each box. Radish sprouts were infested by placing about 200 P. xylostella eggs (24 h old) on 4–5 strips of aluminum egg-laying sheets (3 cm wide, 6 cm long) (see Liang et al., 2003) in each box. To maintain a continuous supply of radish sprouts and P. xylostella larvae, a number of such boxes were prepared and kept in an incubator at 25  $\pm$  2 °C, 50% RH and a photoperiod of 16L:8D.

Pupae of *C. plutellae* were originally obtained from a commercial supplier (Biofac Crop Care, Inc., Mathis, Texas). They were reared under similar laboratory conditions as those used for rearing *P. xylostella*.

#### Insecticides and insecticidal bioassays

Ten insecticides were used in this study, including three azadirachtin-based insecticides, four *Bacillus thuringiensis* (Bt) insecticides, spinosad, and indoxacarb (Table 1). A pyrethroid,  $\lambda$ -cyhalothrin, was used as a commercial standard, and water was used as controls. Concentration for each insecticide used in the bioassay was determined based on the recommended field rate with a delivery rate of 935 l hectare<sup>-1</sup> water. The same concentration for each insecticide was used in all bioassays. Lethal and sublethal effects of these insecticides on pupae and adults of *C. plutellae* were bioassayed. All bioassays were conducted in either plastic or glass petri dishes (9 cm in diameter and 1.5 cm depth).

## Contact toxicity on pupae and adults

All 10 insecticides were used in this bioassay. In each treatment 10 pupae of C. plutellae (2–3 d old) were attached to a glass slide (2 cm  $\times$  7 cm) using

Table 1. Insecticides and concentrations used in this study

Insecticides	Recommended field rate $(ha^{-1})$	Bioassay rate (a.i. 1 <sup>-1</sup> ) <sup>c</sup> Manufacturer	Manufacturer
Agroneem, 0.15% azadirachtin <sup>a</sup> 1.27 g a.i. 1 <sup>-1</sup> 585 ml <sup>b</sup> (8 oz.acre <sup>-1</sup> )  Ecozin, 3.00% azadirachtin 32 g a.i. 1 <sup>-1</sup> 585 ml <sup>b</sup> (8 oz.acre <sup>-1</sup> )  Neemix, 0.25% Azadirachtin 2 g a.i. 1 <sup>-1</sup> 9.3 liter <sup>b</sup> (128 oz acre <sup>-1</sup> )  Dipel (Bt)  Xentari (Bt)  Mattch (Bt)  Crymax (Bt)  SpinTor 2SC spinosad 240 g a.i. 1 <sup>-1</sup> 0.05 kg a.i. (3 oz.acre <sup>-1</sup> )  Avaunt 30% DG Indoxacarb  0.05 kg a.i. (2.5 oz.acre <sup>-1</sup> )	3.5 liter <sup>b</sup> (48 oz.acre <sup>-1</sup> ) 585 ml <sup>b</sup> (8 oz.acre <sup>-1</sup> ) 9.3 liter <sup>b</sup> (128 oz acre <sup>-1</sup> ) 1.12 kg <sup>b</sup> (1 lb.acre <sup>-1</sup> ) 0.05 kg a.i. (3 oz.acre <sup>-1</sup> ) 0.05 kg a.i. (2.5 oz.acre <sup>-1</sup> )	4.8 mg 20.0 mg 1.2 gb 1.2 gb 1.2 gb 1.2 gb 1.2 gb 53 mg	Agro Logistic System, Diamond Bar, CA AMVAC, Los Angeles, CA Certis, Columbia, MD Valent, Walnut Creek, CA Valent, Walnut Creek, CA Dow AgroSciences, Indianapolis, IN Ecogen, Langhorne, PA Dow AgroSciences, Indianapolis, IN Dow AgroSciences, Indianapolis, IN
Warrior 1EC $\lambda$ -cyhalothrin 120 g a.i. $1^{-1}$	$0.026 \text{ kg a.i.} (3 \text{ oz.acre}^{-1}) 28 \text{ mg}$	28 mg	Syngenta, Greensboro, NC

<sup>&</sup>lt;sup>a</sup>1.27 g a.i. per liter (4.8 g per gallon).

<sup>b</sup>Product.

<sup>c</sup>Based on 935 liters of water per hectare (100 gallon per acre).

double-sided sticky tape and were dipped into diluted insecticide solutions or water for ca. 10 s. There were five replicates per treatment. After treatment, the pupae were placed in petri dishes, and adult emergence was recorded. Lethal effects of contact toxicities of the same doses of 10 products used above were evaluated in glass petri dishes (1.5 cm in depth and 9 cm in diameter). The lid and bottom of each petri dish were sprayed with 2 ml of appropriate dilution of each insecticide or water (control) with a potter tower sprayer (Burkard Manufacturing Co. Ltd., Rickmansworth, Hertfordshire, England) at an air pressure of 0.70 kg cm<sup>-2</sup>. The sprayed petri dishes were kept in a fume hood for ca. 1 h, and then six adults (2–3 d old; not separated by gender) were released in each petri dish for 24 h. Each treatment had three replicates. Mortality of *C. plutellae* adults was recorded after 24 h.

# Ingestion toxicity on adult parasitoids

All 10 insecticides were used in this trial at the same rates as in previous tests. *C. plutellae* adults were separately fed with mixtures of insecticide dilutions and 10% honey (vol./vol.), or 10% honey-water solution used as a control. The feeding arenas were made of large plastic petri dishes (1.5 cm in depth and 9 cm in diameter) with a hole (1-cm in diameter) on the side of the petri dish. Two microscopic glass cover slips (1.2 cm  $\times$  1.2 cm) were placed in each petri dish. Sixteen droplets (2  $\mu$ 1 each) of insecticide-honey mixture or honey-water mixture were dispensed on each of the two microscopic slides. Six pairs of *C. plutellae* adults (2–3 d old) were introduced to each petri dish, and they were allowed to feed on the insecticide-honey or honey-water droplets on the microscopic slides. Each treatment had three replicates. Mortality was recorded 24 h after treatment.

To examine acute effects on parasitism, approximately equal numbers of surviving male and female *C. plutellae* adults from each treatment were combined and maintained in a petri dish for mating. Twenty-four h later, 20 second instar *P. xylostella* larvae were offered to each adult female in separate petri dishes. The female adults were allowed to parasitize the larvae. After 10 h of parasitism, female *C. plutellae* adults were removed and exposed *P. xylostella* larvae were reared on radish sprouts. Each treatment had five replicates. Parasitism was assessed based on the number of *P. xylostella* larvae offered and number of *C. plutellae* emerged. Parasitism was not assessed for the treatments if no females survived.

## Persistence of toxicity on adult parasitoids

Three insecticides previously found to be extremely lethal to C. plutellae in glass tests (petri dishes), viz., indoxacarb, spinosad, and  $\lambda$ -cyhalothrin, were

used in this test. Cabbage (variety 'Royal Vantage Hybrid F1') was sown in plastic trays ( $60 \text{ cm} \times 35 \text{ cm} \times 6 \text{ cm}$ ) in a greenhouse on 24 August 2001. The seedlings were transplanted into plastic pots (1.5 liter) on 18 September 2001. The cabbage plants were each sprayed with one of the three insecticides or water until runoff, and placed outside the greenhouse. The plants were watered as needed. Three leaves from each treated plant were removed and a leaf disk of ca. 9 cm in diameter from each leaf was cut out using a scalpel. The leaf disks were placed individually in the petri dishes. Five parasitoid adults (not sexed) were introduced into each petri dish. Each treatment was replicated three times. Mortality was recorded at 24 h. Leaf residues for the three insecticides were bioassayed with parasitoid adults 1, 3, 5, 7 and 10 d after plants were treated.

# Ingestion toxicity for adult longevity

Eight insecticides were used in this test. Each diluted insecticide was mixed with honey (10% v/v). The solution was dispensed to produce 16 droplets (2  $\mu$ l each) on a microscopic glass cover slide. Two microscopic glass cover slips were placed in each petri dish and three pairs of parasitoids (24 h old) were introduced into each petri dish through an opening (1 cm in diameter). Each treatment had three replicates. After 24 h, the parasitoids were removed and maintained in untreated petri dishes where honey-water solution was provided every third day, and longevity of the male and female adults was recorded daily up to the death of all insects.

#### Data analysis

Although the original percentage adult emergence, mortality, parasitism and persistent toxicities were presented in the tables, these data were subjected to arcsin transformation before analysis of variance (one-way ANOVA) through the general linear model (PROC GLM) except for longevity data that were not transformed. Means were separated with Tukey's Studentized Range Test (HSD) at p = 0.05 (SAS Institute, 2001). Percentage parasitism was calculated as: % parasitism = (number of *C. plutellae* pupae/(number of *C. plutellae* pupae + number of *P. xylostella* pupae)) × 100.

*Table 2.* Contact toxicities of selected insecticides on adult emergence and mortality (24 h after emergence at  $25 \pm 2$  °C) of *Cotesia plutellae* when applied to pupae

Insecticides	Concentrations a.i.l <sup>-1</sup>	Emergence $\% \pm SE^{ab}$	Mortality $\% \pm \mathrm{SE}^{\mathrm{ab}}$
Neemix	20 mg	$90.0 \pm 5.7 \text{ a}$	$16.7 \pm 9.6 \mathrm{c}$
Agroneem	4.8 mg	$73.3 \pm 6.6 \text{ a}$	$11.1 \pm 11.1 \text{ c}$
Ecozin	20 mg	$90.0 \pm 10.0 \text{ a}$	$5.6 \pm 5.6 \text{ b}$
Xentari	1.2 g	$63.3 \pm 8.8 \text{ a}$	$0.0 \pm 0.0$ a
Mattch	1.2 g	$66.6 \pm 8.8 \text{ a}$	$5.6 \pm 5.6 \text{ b}$
Crymax	1.2 g	$76.6 \pm 8.8 \; a$	$0.0 \pm 0.0$ a
Dipel	1.2 g	$70.0 \pm 5.7 \text{ a}$	$11.1 \pm 5.6 \text{ c}$
Spinosad	53 mg	$73.3 \pm 6.6 \text{ a}$	$50.0 \pm 9.6 c$
Indoxacarb	53 mg	$73.3 \pm 8.8 \text{ a}$	$100.0 \pm 0.0 \; \mathrm{d}$
λ-cyhalothrin	28 mg	$76.6 \pm 3.3 \text{ a}$	$88.9 \pm 5.6 \text{ d}$
Control	_	$90.0 \pm 5.7 \text{ a}$	$0.0 \pm 0.0$ a

<sup>&</sup>lt;sup>a</sup>Means ( $\pm$  SE) within a column followed by the same letter are not significantly different at p < 0.05; Tukey's Studentized Range Test (HSD).

#### **Results**

## Contact toxicity on pupae and adults

Toxic effects of field dosages of all 10 insecticides on *C. plutellae* pupae were relatively low, and the percentages of adult emergence were not significantly different among all insecticides or with the water control (Table 2). However, mortality of adults 24 h after emergence was significantly different among insecticides. Compared with water, the four Bt-insecticides had no or little acute toxic effect on *C. plutellae* adults emerging from treated pupae, with no effects by Xentari and Crymax, 5.6% mortality by Mattch, and 11.1% mortality by Dipel. Among the azadirachtin-based insecticides, Ecozin was significantly less toxic (5.6%) to *C. plutellae* adults than Neemix (16.7%) and Agroneem (11.1%). In contrast, the greatest adult mortality occurred among those emerged from pupae treated with indoxacarb (100%) and  $\lambda$ -cyhalothrin (88.9%).

<sup>&</sup>lt;sup>b</sup>The emergence and mortality (%) are untransformed data while the data used in the analysis were arcsin-transformed.

*Table 3.* Effects of ingestion toxicities of 10 insecticides on the mortalities and parasitism rates of *Cotesia plutellae* (after 24 h exposure at  $25 \pm 2$  °C)

Insecticides	Concentration a.i.l <sup>-1</sup>	Mortality $\% \pm SE^{ab}$	Parasitism % ± SE <sup>ab</sup>
Neemix	20 mg	$0.0 \pm 0.0$ a	$37.6 \pm 3.9 \text{ c}$
Agroneem	4.8 mg	$0.0 \pm 0.0$ a	$40.1 \pm 4.4 \text{ c}$
Ecozin	20 mg	$0.0 \pm 0.0$ a	$44.3 \pm 3.5 \text{ c}$
Xentari	1.2 g	$0.0 \pm 0.0$ a	$66.2 \pm 5.2 \text{ b}$
Mattch	1.2 g	$0.0 \pm 0.0$ a	$69.5 \pm 6.1 \text{ ab}$
Crymax	1.2 g	$5.6 \pm 5.6$ a	$80.9 \pm 3.7 \text{ ab}$
Dipel	1.2 g	$0.0 \pm 0.0$ a	$77.4 \pm 3.5 \text{ ab}$
Spinosad	53 mg	$100.0 \pm 0.0 \text{ b}$	_
Indoxacarb	53 mg	$11.1 \pm 5.6 \text{ a}$	_
λ-cyhalothrin	28 mg	$100.0 \pm 0.0 \text{ b}$	_
Control	_	$0.0 \pm 0.0$ a	$88.1 \pm 3.2 \text{ a}$

<sup>&</sup>lt;sup>a</sup>Means ( $\pm$  SE) within a column followed by the same letter are not significantly different at p < 0.05; Tukey's Studentized Range Test (HSD).

## Ingestion toxicity on adult parasitoids

All adults died after ingestion of spinosad and  $\lambda$ -cyhalothrin. In contrast, no adult mortality was recorded in the treatments of Neemix, Agroneem, Ecozin, Xentari, Mattch, and Dipel, whereas only 5.6% and 11.1% mortalities were found in the treatments of Crymax and indoxacarb, respectively. All insecticidal treatments reduced the percentage parasitism of the surviving females as compared with the water control (Table 3).

# Persistence of toxicity on adult parasitoids

Effects of leaf residues of  $\lambda$ -cyhalothrin, indoxacarb and spinosad on C. plutellae adults varied significantly (Table 4).  $\lambda$ -cyhalothrin caused the highest mortality among the three insecticides in the first 3 days after treatment with 100% mortality compared with 56% mortality by indoxacarb and 22% mortality by spinosad. The leaf residues of  $\lambda$ -cyhalothrin and indoxacarb lasted longer than that of spinosad. By day 10, the leaf residue of  $\lambda$ -cyhalothrin still caused 39% mortality, and that of indoxacarb caused 44% mortality. In contrast, the leaf residue of spinosad lasted <10 days with no effects on C. plutellae on the 10th day.

<sup>&</sup>lt;sup>b</sup>The mortality and parasitism (%) are untransformed data while the data used in the analysis were arcsin-transformed.

*Table 4.* Contact effects of three selected insecticides on mortality of *Cotesia plutellae* adults exposed to residue at various intervals after treatment of cabbage (after 24 h exposure at  $25 \pm 2$  °C)

	Concentra-	Mortality % ±	: SE <sup>ab</sup>			
Insecticides	tion a.i.l <sup>-1</sup>	1 d	3 d	5 d	7 d	10 d
λ-cyhalothrin	28 mg	$100.0 \pm 0.0 \text{ c}$	$100.0 \pm 0.0  \mathrm{d}$	$61.1 \pm 5.6 \mathrm{c}$	$38.9 \pm 5.6 \text{ ab}$	$38.9 \pm 5.6  \mathrm{b}$
Spinosad	53 mg	$38.9 \pm 5.6 \text{ b}$	$22.2\pm5.6~\mathrm{b}$	$22.2 \pm 5.6 \text{ ab}$	$24.0 \pm 5.6\mathrm{b}$	$0.0 \pm 0.0$ a
Indoxacarb	53 mg	$50.0 \pm 9.6 \text{ b}$	$55.6 \pm 5.6~\mathrm{c}$	$50.0 \pm 9.6~bc$	$44.4\pm11.1\;c$	$44.1\pm11.1\;b$
Control	_	$0.0 \pm 0.0$ a	$0.0 \pm 0.0$ a	$0.0\pm0.0~a$	$0.0\pm0.0~\mathrm{a}$	$0.0\pm0.0~\mathrm{a}$

<sup>&</sup>lt;sup>a</sup>Means ( $\pm$  SE) within a column followed by the same letters are not significantly different at p < 0.05; Tukey's Studentized Range Test (HSD).

Table 5. Effects of ingestion toxicities of eight pesticides on gender longevity of Cotesia plutellae

Concentration	Gender longevity days $\pm$ SE <sup>a</sup>		
$a.i.l^{-1}$	Male	Female	
20 mg	$10.7 \pm 0.3 \text{ a}$	$12.7 \pm 0.3 \text{ ab}$	
4.8 mg	$10.0 \pm 0.6 \text{ a}$	$11.7 \pm 0.3$ ab	
20 mg	$9.7 \pm 0.7 \text{ a}$	$12.0 \pm 0.6$ ab	
1.2 g	$11.0 \pm 0.6 \text{ a}$	$12.7\pm0.7~ab$	
1.2 g	$10.7 \pm 0.7 \text{ a}$	$12.3 \pm 0.7 \text{ ab}$	
1.2 g	$7.0 \pm 3.0 \text{ a}$	$12.7\pm0.7~ab$	
1.2 g	$9.6 \pm 0.7 \; a$	$11.7 \pm 1.2$ ab	
53 mg	$5.6\pm2.3~a$	$7.7 \pm 3.4 \text{ b}$	
_	$11.7 \pm 0.9 a$	$14.3 \pm 0.7 \text{ a}$	
	a.i.l <sup>-1</sup> 20 mg 4.8 mg 20 mg 1.2 g 1.2 g 1.2 g 1.2 g	a.i.l <sup>-1</sup> Male       20 mg $10.7 \pm 0.3$ a       4.8 mg $10.0 \pm 0.6$ a       20 mg $9.7 \pm 0.7$ a       1.2 g $11.0 \pm 0.6$ a       1.2 g $10.7 \pm 0.7$ a       1.2 g $7.0 \pm 3.0$ a       1.2 g $9.6 \pm 0.7$ a       53 mg $5.6 \pm 2.3$ a	

<sup>&</sup>lt;sup>a</sup>Means ( $\pm$  SE) within a column followed by the same letter are not significantly different at p < 0.05; Tukey's Studentized Range Test (HSD).

## Ingestion toxicity for adults longevity

No significant differences were detected in male longevity among the insecticide treatments as compared with that in the water control (Table 5). Although the longevity of the female adults of *C. plutellae* (7.7 d) in the indoxacarb treatment was significantly shorter than that in the water control (14.3 d), it was not significantly different from those in other insecticide treatments. These results indicate that all these insecticides had little or no significant effects on the longevity of *C. plutellae* adults.

<sup>&</sup>lt;sup>b</sup>The mortality and parasitism (%) are untransformed data while the data used in the analysis were arcsin-transformed.

#### **Discussion**

Our results showed that the effects of an insecticide on *C. plutellae* varied significantly depending on the stages of parasitoid treated and the insecticides tested. Although the three neem- or azadirachtin insecticides caused relatively low mortality of *C. plutellae*, conflicting results on effects of neem- or azadirachtin-based insecticides on this species can be found in the literature, from harmless to detrimental. For example, Mani (1995) found that a neem seed kernel extract and a commercial neem insecticide, Neemark, were harmless to *C. plutellae* adults. In contrast, Perera et al. (2000) reported that two neem seed kernel extracts were detrimental to emergence of *C. plutellae*. Verkerk and Wright (1994) also found that two neem insecticides significantly reduced adult emergence from treated cocoons. We strongly recommend testing the efficacy of azadirachtin-based insecticides of the target pest and beneficial arthropods before they are used because of the dramatic differences among the commercial insecticides in formulation, active ingredient and non-active ingredients.

Our results also showed that all four Bt insecticides had no effects on C. plutellae adults and pupae. These results were similar to other reports in the literature. Bt-insecticides had no effect on longevity and oviposition of C. plutellae adults (Chilcutt and Tabashnik, 1999). Kao and Tzeng (1992) reported that two Bt insecticides (Dipel and SAN 415) had no effects on C. plutellae adults, but slightly reduced parasitism. Normally, Bt-insecticides had no significant direct effect on C. plutellae under field conditions (Uematsu and Yamashita, 1999). However, under field conditions, effects of Bt-based insecticides on C. plutellae or other parasitoids may be influenced by factors, such as the age and density of the host. Loganathan et al. (2001) reported that although Bt insecticides were generally not detrimental to C. plutellae, in one test they found that the C. plutellae population was significantly higher in untreated control (3.2–4.7 pupae/plant) when compared to the treatments with a Bt insecticide, Spicturin (0.2-1.8 pupae/plant), and they thought that these effects were due to the lower population of the parasitoid in the Spicturin treatments associated with the reduction of larval population of *P. xylostella*.

Indoxacarb was designated as a reduced-risk product by the Environmental Protection Agency of the United States (Anonymous, 1998). It has been reported that indoxacarb has no detrimental effects on hemipteran predators (*Geocoris* spp., *Orius* spp., and *Nabis* spp.), hymenopterous parasitoids (*Aphidius* sp., *Cotesia* sp., *Bracon* spp., *Microplitis* spp., and *Trichogramma* sp.), spiders and predacious mites, and has little or no adverse effects on lacewings [*Chrysoperla rufilabris* (Burmeister)] and coccinellids (Ruberson and Tillman, 1999; Studebaker and Kring, 1999). However, results

from this study clearly show that laboratory-aged leaf residue of indoxacarb can be significantly toxic to adults of *C. plutellae*.

Effects of spinosad on *C. plutellae* from this study were similar to those reported in the literature. Pietrantonio and Benedict (1999) and Ruberson and Tillman (1999) found that spinosad residue on cotton leaves was toxic to *C. plutellae* and *Cotesia marginiventris* (Cresson), respectively, and highly toxic to *Trichogramma pretiosum* Riley. Tillman et al. (1998) found that when directly sprayed on beneficial insects, spinosad at lower rates (0.05–0.07 kg AI ha<sup>-1</sup>) resulted in high to moderately high survival of *Cardiochiles nigriceps* Viereck, *Coccinella setempunctata* L. and *Geocoris punctipes* (Say), but the highest rate (0.1 kg AI ha<sup>-1</sup>) was very toxic to *G. punctipes* and *C. marginiventris*.

Haseeb and Amano (2002) recorded reduction in parasitism when some insect growth regulators (chlorfluazuron, flufenoxuron and tefludenzuron) were ingested by C. plutellae adults. This may suggest that these insecticides may in some way affect oogenesis in parasitoids and the development of eggs laid inside host larvae. Parasitoid behavior is another parameter that could be altered by the application of certain conventional insecticides, affecting parasitism rates. Avoiding insecticide use at times when adult parasitoids are present can reduce mortality. We recommend further testing of azadirachtinbased and Bt insecticides under field conditions, especially with respect to parasitoid behavior and physiology. Earlier insecticide testing schemes relied on residual exposure to dry deposits (Hassan, 1994), often on inert surfaces for an extended period, thus ensuring exposure and uptake. These tests, however, generally avoid direct spray and uptake from contaminated prey or treated host or treated food plants (especially nectar feeding adult parasitoids). These other routes of uptake may be important to some beneficial arthropods (Jepson, 1989) and certain novel insecticides such as Bt- and azadirachtin-based insecticides may require dietary uptake in order to show toxic effects.

Standard methods to test the side-effects of pesticides on numerous natural enemies of insects and mites have been developed by the International Organization of Biological Control (IOBC) (Hassan et al., 1985), however, no standard method has been developed for *C. plutellae*. We followed the general principle and procedures by the IOBC, and developed the technique used in this study and other previous studies (Haseeb, 2001; Haseeb et al., 2001). We think that this technique is simple, and provides reliable information on the adverse effects of these insecticides on *C. plutellae*. Although laboratory tests give repeatable and predictable levels of exposures, they cannot realistically reproduce exposure in the field. Brown et al. (1990) recommended that a combination of direct spraying and residual contact with fresh deposits was

the most appropriate combination for pesticide testing against certain groups of beneficial organisms. We also recommend that similar studies may be undertaken with other standard test organisms to optimize the presentation of the insecticides and the number of routes by which exposure might occur.

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